Helicobacter pylori gastritis and primary gastric non-Hodgkin's lymphomas

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Abstract
Aims—To evaluate further the relation between gastric malignant lymphoma of the mucosa associated lymphoid tissue (MALT) and Helicobacter pylori.
Methods—One hundred and sixty two surgical specimens of MALT lymphoma were retrospectively investigated to determine tumour type and inflammatory patterns. In 121 cases biopsy specimens obtained before surgery were available and stained with haematoxylin and eosin, periodic acid Schiff, Giemsa and Warthin-Starry stains.
Results—Residual lymphoid follicles were found less often in high grade malignant than in low grade malignant MALT lymphomas. Chronic active gastritis was shown within the mucosa at some distance from the tumours in 143 of 146 specimens. In all the cases for which biopsy specimens could be evaluated, colonisation of the mucosa by H pylori had occurred. Lymphoid follicles and lymphoid aggregates were detected in 82.7% of the antral, and in 85% of the body mucosa specimens.
Conclusions—These data support the hypothesis that H pylori has an important role in the development of MALT lymphomas. Furthermore, the chronic inflammation preceding malignant transformation might enhance the probability of malignant transformation via chronic stimulation of the lymphoid tissue. This might in part indicate why MALT lymphomas occur most often in the stomach.

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One of the most exciting current developments in gastroenterology is the discovery that Helicobacter pylori has a decisive role in gastroduodenal disease.1-3 Recently, several groups have published data showing a correlation between H pylori and gastric carcinomas.4-4 Wotherspoon et al were the first to investigate the presence of H pylori in larger numbers of gastric lymphomas of the mucosa associated lymphoid tissue (MALT lymphomas): in 92% of cases H pylori was indeed detected.7 On the basis of earlier observations that the prevalence of lymphoid follicles correlated significantly with the detection of H pylori,8-8 they suggested that H pylori might trigger the acquisition of MALT in the gastric mucosa which usually contains only a few T lymphocytes.10 In the light of these findings H pylori and the subsequent inflammatory response might be prerequisites for the development of a MALT lymphoma in the stomach. Further support for this hypothesis was provided by data published by Doglioni and coworkers which showed that the high prevalence of gastric MALT lymphoma correlated closely with a high prevalence of H pylori infection in a town in northern Italy.11 If the suggested route to MALT through H pylori infection to the malignant transformation holds true, then not only H pylori but an inflammatory response to H pylori infection should be evident in almost all cases of MALT lymphoma as well. To elucidate further this association we retrospectively investigated 162 cases of gastric resection specimens for gastric lymphoma.

Methods
One hundred and sixty six surgical specimens of gastric lymphomas (56 gastrectomies, 95 arboral resections, nine local excisions, six resections of the gastric remnant were examined at the Institute of Pathology, Bayreuth, over a period of 11 years. They were fixed in 10% formalin, embedded in paraffin wax and routinely stained with haematoxylin and eosin periodic acid Schiff and Giemsa. The lymphomas were classified according to the proposal of the European Lymphoma Study Group.12 Only B cell lymphomas of MALT type were included in this study. Two plasmacytomas and two T cell lymphomas were excluded, reducing the number of cases investigated to 162.

The phenotypic evaluation of lymphomas was done using monoclonal antibodies recognising formalin fixation resistant antigens (L26, UCHL1, MT1, all from Dako) and a subsequent peroxidase-antiperoxidase method. The high grade malignant MALT lymphomas were divided into those with and without a low grade component. Furthermore, the depth of infiltration into the gastric wall was evaluated using the International Union against Cancer classification of gastric cancer (table 1).13 Clinical staging of lymphoma was also carried out in all patients using abdominal computed tomograms or ultrasonography, liver and bone marrow biopsies

| Table 1: UICC classification of gastric carcinoma |
|---|---|
| pT1 | Tumour restricted to mucosa and submucosa |
| pT2 | Tumour infiltrating beyond the submucosa into the muscularis propria and the subserosal layer |
| pT3 | Tumour penetrating the serosa |
| pT4 | Tumour continuously infiltrating other organs (liver, spleen) |
with subsequent classification according to the modified Ann Arbor scheme\textsuperscript{14}\textsuperscript{-15} (table 2), and only tumours in which the largest tumour mass was located in the stomach. The presence of non-neoplastic reactive lymphoid follicles within the tumours was registered.

In 146 cases it was also possible to evaluate the gastric mucosa at least 4 cm away from the tumours for infiltration of the lamina propria with lymphocytes, plasma cells, and neutrophilic polymorphs as well as intestinal metaplasia. The prevalence of lymphoid aggregates and follicles was also registered. The latter were diagnosed when germinal centres could be detected. \textit{H pylori} gastritis was diagnosed when a lymphoplasmacellular infiltrate was combined with neutrophilic polymorphs concentrated around the neck region with infiltration of gastric glands. In accordance with these criteria, all cases of chronic active gastritis were classified as \textit{H pylori} gastritis. Because of the influence of delayed fixation on the detection of \textit{H pylori} (M Stolte, unpublished observations) we also investigated endoscopic biopsy specimens taken three months or less before the surgical procedure for colonisation of the gastric mucosa with \textit{H pylori}; such biopsy specimens were available in 121 cases. \textit{H pylori} was detected using the Warthin-Starkey stain. Lymphocytic and reactive gastritis were diagnosed according to the criteria given before.\textsuperscript{15}\textsuperscript{-17}

Statistical evaluation was done using the \( \chi^2 \) test.

**Results**

The mean age of the patients (74 men, 88 women) was 62.2 years, with a standard deviation of 13.1 years and a range from 20 to 92 years. The types of lymphomas detected, together with their prevalences, are shown in table 3. The staging of the tumours according to the Ann Arbor classification showed that most were only locally infiltrating: E I, in 72 (44.4\%) cases and B I, in 33 (20.4\%) cases. Only in 41 (25.3\%) patients was an infiltration of regional lymph nodes detected (E II), and in four (2.5\%) cases had infiltrated distant lymph nodes. Liver or spleen metastases were found in three (1.9\%) cases and bone marrow infiltration in two (1.2\%). In seven (4.3\%) patients in whom only a local excision was performed no classification was possible. The correlation between the depth of infiltration and the grade of the lymphomas (table 4) reached significance (p < 0.001). The largest tumour diameters are shown in table 5. The types of gastritis detected in the mucosa located at a distance from the tumours are listed in table 6. In all the cases of chronic active gastritis the biopsy specimens taken before surgery showed \textit{H pylori} colonisation of the gastric mucosa. A correlation between largest diameters, grade, or depth of infiltration could not be demonstrated. Reactive non-neoplastic lymphoid follicles were found within the tumours in 62.0\% of cases. An analysis of their correlation with degree of malignancy and with depth of infiltration reached significance in the case of the former (p < 0.001) (table 7). In the latter, however, owing to the small number of cases in the pT4 group, statistical analysis was possible only by pooling pT3 and pT4 lesions (p < 0.05) (table 8). The prevalence of lymphoid follicles and lymphoid aggregates in the antral and gastritis were diagnosed according to the criteria given before.\textsuperscript{15}\textsuperscript{-17}

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body mucosa is shown in table 9. The distribution of intestinal metaplasia of the gastric mucosa located at a distance from the lymphomas is shown in table 10. Significant correlations with the degree of malignancy or the depth of infiltration could not be shown.

Discussion
A MALT system as exemplified in Peyer’s patches is not present in the normal gastric mucosa. Following infection with H pylori, MALT is acquired. Further evidence for this sequence of events is provided by the results of eradication studies. After successful eradication of H pylori, the density of the lymphoplasmacellular infiltration in the lamina propria decreased substantially and lymphoid aggregates and lymphoid follicles, present before were no longer detectable in control biopsy specimens (unpublished own observations). More support for the hypothesis that H pylori plays an important part in the genesis of MALT lymphoma comes from recent serological, epidemiological, and histopathological research. Parsonnet et al reported in 1991 that in patients with gastric lymphomas antibodies against H pylori were present in 90.9% compared with 63.6% of matched controls. They only investigated 11 patients. Wotherspoon et al histologically evaluated 110 MALT lymphomas of the stomach in both biopsy and surgical specimens, looking for the presence of H pylori, and found the organisms in 92% of cases. This prevalence of H pylori was significantly higher than that reported for non-ulcer dyspepsia (50–60%). An analysis of epidemiological data published by Doglioni et al showed a high prevalence of primary gastric lymphoma in a town in northern Italy, correlated with a high prevalence of H pylori infection. Similarly, a low prevalence of gastric lymphoma was observed in a number of British towns with a low prevalence of H pylori.

The data presented here are based exclusively on findings in surgical specimens, and for the first time the gastritis associated with the lymphomas has been investigated as well. An extremely high prevalence of chronic active gastritis with features typical of H pylori aetiology was demonstrated in gastric MALT lymphoma (97.7%), compared with the results of a study of 5000 gastric biopsy specimens of various forms of gastritis at our insti-
infection can trigger lymphoproliferation has been shown in Epstein-Barr infection.27 28 If the assumption is correct that H pylori infection is not merely the basis for, but actively contributes to, the aetiology of gastric lymphoma, then in view of the far greater prevalence of H pylori infection than of MALT lymphoma, additional factors must operate. These might include peculiarities of the H pylori strain involved: the high prevalence of lymphoid follicles and lymphoid aggregates might be interpreted as features of a possible high immunogenicity. Furthermore, exogenic—for example, dietary factors, virus infections, or disorders of immune regulation—have to be taken into consideration. The chronic inflammation caused by H pylori could be an important part of the answer to the question why the stomach has the highest prevalence of MALT lymphomas,29-31 although in the normal stomach only a few lymphocytes are found in the lamina propria and epithelium.10 Studies aimed at analysing the additional factors mentioned and estimating the possible benefit of eradication of H pylori are in progress.

Although many questions concerning MALT lymphomas remain to be elucidated, we conclude that our results, together with the reported data, strongly support the hypothesis that H pylori gastritis, by leading to the acquisition of MALT, is a prerequisite for the development of a MALT lymphoma in the stomach in almost all cases. Whether H pylori and the inflammatory response also actively contribute to the formation of lymphomas in the stomach has yet to be determined.

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References